Space Sciences Laboratory
University of California U., Berkeley.
Berkeley 4, California

BIOCHEMICAL ACTIVITIES OF TERRESTRIAL MICROORGANISMS IN SIMULATED PLANETARY ENVIRONMENTS

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Samuel Silver

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Introduction:

Current efforts continue to be directed toward the study of environmental parameters which relate directly or indirectly to the problem of biological contamination and indigenous life on Mars. Recent results, reports and papers being prepared for publication, and work in progress are briefly discussed.

1. A paper entitled "Biological contamination of Mars: survival of terrestrial microorganisms in simulated Martian environments" by Stanley Scher, Elliot Packer and Carl Sagan was presented by Dr. Scher at the Fourth International Space Science Symposium in Warsaw. The paper calls attention to the relatively rigorous conditions which prevail on the Martian surface which have led some authors to doubt the likelihood of biological contamination or indigenous life on Mars. Experiments were reported which indicate that every sample of terrestrial soil tested contains a small fraction of microorganisms which survive under simulated Martian conditions.

The demonstration that terrestrial microorganisms can survive on Mars, has, of course, no rigorous implications for the existence of life on that planet; however, these experiments do provide evidence that biological mechanisms exist for survival under these conditions and suggest that indigenous organisms evolving on Mars may have developed the capability for dealing with the apparent rigors of the Martian environment. The implication of these results for the question of biological contamination of Mars is clear. The survival of terrestrial microorganisms in simulated Martian conditions underscores the necessity for scrupulous sterilization of all spacecraft intended for Mars landings.

An abstract of the paper is appended and reprints will be forwarded to NASA when they become available.

2. A second paper concerned with the biological contamination problem is being prepared for publication in Icarus. This work considers low temperatures and aridity as constraints on the survival and growth of terrestrial microorganisms in simulated planetary environments. Critical factors in microbial survival and growth at low temperatures are reviewed. The paper also includes some discussion of approaches to the design of planetary simulation experiments and certain photochemical aspects of Martian ecology.

3. We have conducted additional experiments in which microorganisms derived from soil were exposed to temperatures in excess of 100°C. These studies relate to both microbial survival on the lunar surface and to the adequacy of existing sterilization recommendations for spacecraft on planetary missions.

For sterilization of impacting vehicles, a 24 hour dry heat cycle at 130-140°C. has been suggested. Since this procedure is claimed to lower the microbial spore population by a factor of 10^{-13}, the question arose: Do survivors of such heat treatments also survive the low temperatures and rigors of the Martian environment? The results of experiments to date indicate that microorganisms derived from terrestrial soil were unable to tolerate exposure to 130-140°C. for 24 hours, and suggests that the current procedures for spacecraft sterilization are adequate if properly applied.

4. Studies on the lability of the photosynthetic apparatus in algal flagellates following exposure to low levels of ultraviolet irradiation or growth at supra-optimal temperatures are in progress. To follow the kinetics of inactivation and recovery of the ultraviolet sensitive sites which control the heredity of the chloroplast, we are comparing the uv dose required to yield a given number of permanently colorless (non-photosynthetic) cells during the course of exposure to environmental temperatures which selectively inhibit chloroplast formation. The results of preliminary experiments suggest that this approach may provide an accurate index of the number of uv sensitive sites required to maintain the hereditary character for chloroplast-forming ability in cells undergoing treatments which lead to loss of a functional photosynthetic apparatus.

To obtain evidence in support of the nucleoprotein character of the uv sensitive sites, we are attempting to incorporate pyrimidine analogs into the nucleic acid fraction of Euglena. The radiation sensitivity and other properties of halogenated nucleic acids offer a number of exciting approaches to this problem. Since the methyl group of thymine can be replaced by halogens having similar van de Waals radii or partial specific volumes, cells grown under conditions of thymine deficiency might be expected to substitute 5-bromouracil for thymine. To effect maximum incorporation, cells are induced to utilize exogenous thymine by means of drugs such as sulfanilamide which are known to reduce thymine synthesis by interference with folic acid metabolism. In a preliminary study we have observed a tenfold increase in

cells grown in the presence of bromouracil under conditions which impair thymine synthesis. Subsequent experiments will be carried out to verify these results and to establish the incorporation of bromouracil.

5. A paper entitled "On the nature of the Jovian Red Spot" by Carl Sagan was published in the Proceedings of the 11th International Astrophysics Colloquium in Liege. The paper suggests that the color of the Red Spot is due to organic matter characteristic of considerable depths in the Jovian atmosphere being convectively transported to the top of the atmosphere. Reprints will be sent under separate cover.

BIOLOGICAL CONTAMINATION OF MARS: SURVIVAL OF TERRESTRIAL MICROORGANISMS IN SIMULATED MARTIAN ENVIRONMENTS

(abstract)

S. Scher, E. Packer, and C. Sagan*

Space Sciences Laboratory, University of California
Berkeley, California, U. S. A.

It has been postulated that the accidental introduction of terrestrial microorganisms to other planets during the course of space exploration might impede or bias the detection of organic matter and possible indigenous organisms, and thereby confuse subsequent studies of extraterrestrial life. To assess the likelihood of biological contamination of Mars, we have applied the principle of natural selection on a laboratory scale. Terrestrial microorganisms were collected from a variety of environments, including regions of high alkalinity, low mean daily temperature, and low annual rainfall. The air-dried soils were then subjected to a simulated Martain environment involving 12-hour freeze-thaw cycles from about -60°C to about +20°C; atmospheres of 95 per cent nitrogen, 5 per cent carbon dioxide and low moisture content; $\leq$ 0.1 atm pressure; and a total ultraviolet dose at 2537 Å of 10^9 erg cm^{-2}. In some experiments, organic supplements were provided. Survivors were scored on supplemented agar. Preliminary results indicate a wide variety of survivors, even when no organic supplements were introduced. Survivors included obligate and facultative anaerobic spore-formers and non-spore-forming facultative anaerobic bacteria. Diurnal freezing and thawing was continued for six months. There was no significant loss of viability after the first freeze-thaw cycle. An extensive literature survey shows that survival of terrestrial microorganisms under individual simulated Martian conditions has been known for decades. The present investigation shows the absence of pronounced synergistic effects inhibiting survival. The probable existence of organic matter and moisture on Mars, at least in restricted locales and times, makes it especially likely that terrestrial microorganisms can also reproduce on Mars. The demonstration that all samples of terrestrial soil tested contain a population of microorganisms which survive in simulated Martian environments strongly underscores the need for scrupulous sterilization of all spacecraft intended for Mars landing.

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*Permanent address: Department of Astronomy, Harvard University; and Smithsonian Astrophysical Observatory, Cambridge, Massachusetts, U. S. A.